supplying altered peptide ligands, developing tolerance by i.v. and oral administration, and blocking costimulatory molecules (Sakai K. et al., Proc. Natl. Acad. Sci. U.S.A. 86:9470 (1989); Hurtenbach U. et al., J. Exp. Med.. 177:1499 (1993); Fairchild P.J. et al., Immunology 81:487 (1994); Brocke S. et al., Nature 379:343 (1996)).

However, there is no cure for MS, a disease which is ultimately fatal. There is a need for improved agents to treat MS and other demyelinating conditions.

Brief Description of the Drawings

FIG. 1 is a line graph that shows the inhibition of biotinylated Cop 1 (FIG. 1A) and MBP 85-99 (FIG. 1B; SEQ ID NO: 1) binding to HLA-DR2 molecules as a function of concentration of unlabeled Cop 1 or each of the synthetic peptides: MBP 85-99 (SEQ ID NO: 1); and #100-#103 (SEQ ID NOs: 64-67, respectively).

FIG. 2 is a line graph that shows the inhibition of proliferation of HLA-DR2 restricted MBP 84-102-specific T cell line transfectants Hy1B (FIG.2A) and 8073 (FIG.2B) as a function of the concentration of Cop 1 or each of synthetic peptides: #94 (SEQ ID NO: 60); #96 (SEQ ID NO: 62); #99 (SEQ ID NO: 63); #100-#103 (SEQ ID NOs: 64-67, respectively); and #107 (SEQ ID NO: 69).

FIG. 3A on the left is a line graph that shows the inhibition of biotinylated MBP 85-99 by each of MBP 85-99 (SEQ ID NO: 1), or #101 (designated #1 in the FIG., solid triangles; SEQ ID NO: 65); FIG. 3A on the right is a line graph that shows inhibition of biotinylated MBP 85-99 binding to HLA-DR2 molecules as a function of concentration of synthetic peptide #101 (designated #1 in the FIG., solid triangles; SEQ ID NO: 65); #4 (SEQ ID NO: 92); #6 (SEQ ID NO: 94); and #7 (SEQ ID NO: 95). FIG. 3B is a line graph that shows the inhibition of proliferation of HLA-DR2 restricted MBP 84-102-specific T cell transfectant 8073 as a function of the concentration each of synthetic peptides: #101 (designated #1 in the FIG., solid triangles; SEQ ID NO: 65); and #4 (SEQ ID NO: 92).

FIG. 4A is a line graph that shows the inhibition of biotinylated MBP 85-99 binding to HLA-DR2 molecules as a function of concentration of each of unlabeled peptides MBP 85-99 (SEQ ID NO: 1); #2 (SEQ ID NO: 85); #3 (SEQ ID NO: 91); and #5 (SEQ ID NO: 93). FIG. 4B is a bar graph comparing the data obtained for these peptides at a concentration of 1.3 μM, and shows that for peptides that are otherwise identical in sequence, hydrophobic residues at the P1 position that are less bulky, such as valine (V) result in a peptide that is more inhibitory, compared to residues that are more bulky such as tyrosine (Y). FIG. 5A is a bar graph showing data obtained for inhibition of binding of biotinylated MBP 85-99 to HLA-DR2 molecules peptides at a concentration of 1.3 μM for each of unlabeled peptides MBP 85-99

BSB 4/6/05

human immunodominant epitope of MBP, to HLA-DR2 (DRB1*1501) molecules (Fridkis-Hareli M, et al., J Immunol 160:4386-4397, 1998). Copolymers composed of only three amino acids (for exampke, EAK, YEA, YAK and YEK) also bind to purified HLA-DR1, -DR2 and -DR4 molecules (Fridkis-Hareli M, et al. Int Immunol 11:635, 1999; PCT/US99/16,617). Moreover, these three amino acid copolymers compete with CII 261-273 for binding to RA-associated HLA-DR1 (DRB1*0101) and -DR4 (DRB1*0401) molecules, and also inhibited CII-reactive T cell clones (Fridkis-Hareli M, et al. Proc Natl Acad Sci USA 95:12528, 1998); PCT/US99/16617 and PCT/US99/16747.

The bound fraction of Cop 1, treated with aminopeptidase I, has been isolated from recombinant "empty" HLA-DR molecules produced in insect cells, and has been sequenced. The Cop 1 binding motif for HLA-DR2 showed increases in levels of E at the first and second cycles, of K at the second and third cycles, and of Y and A (presumably at P1 of the bound peptide) at the third to fifth cycle. No preference was seen at the following cycles which were mainly A (Fridkis-Hareli M, et al. J Immunol 162:4697, 1999; PCT/US99/16,617). Recently, the characterization of the active component(s) of the mixture of random polypeptides was attempted by synthesis of a set of peptides based on Cop 1 binding properties to HLA-DR1 and -DR4 molecules (Fridkis-Hareli M, et al. Human Immunol 61: 640, 2000); PCT/US99/16,617. Several peptides inhibited binding of CII 261-273 epitope to DRB1*0101 and -DR4 DRB1*0401 molecules and inhibited presentation of this epitope to CII-reactive DR1- and DR4-restricted T cell clones (Fridkis-Hareli M, et al. Human Immunol 61: 640, 2000).

Demyelinating conditions have been found to occur post-viral infection, post-vaccination, post-encephalomyelitis (Wucherpfennig K.W. et al., Immunol. Today 12:277-282 (1991)) and following administration of certain anti-TNF agents (*FDA Talk Paper*, Food and Drug Administration Public Health Service, Rockville, MD). ~~http://www.fda.gov/bbs/topics/ANSWERS/ANSDO954.html).~~

BSB 4/16/05

Many derivatives of synthetic peptides having increased pharmacological life in vivo have been synthesized. The loading of MHC class II binding sites occurs in endosomal compartments abundant with proteases, particularly cathepsins. Peptides may be digested also by amino- or carboxy- peptidases in serum or other biological fluids. Therefore, proteolysis of the peptides may effectively remove the peptides from the subject (Bennett, K., et al., 1992, Eur. J. Immunol. 22:1519). To reduce or eliminate potential proteolysis, modification of the peptides, for example, N- methylation of backbone nitrogens in the peptides, which are not involved in essential hydrogen bonding interactions, could produce a peptide derivative that is resistant to proteolysis (Falconi, F., et al., 1999, Nature Biotechnology 17:562). In Falconi et